Supplementary Information

Toehold-mediated DNA logic gates based on host-guest DNA-GNPs

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Materials

Oligonucleotides were synthesized by Shanghai Sangon Biological Engineering Technology &Services CO.,Ltd.. All other reagents were of analytical reagent grade and used without further purification. Ultrapure water was used throughout the experiments. Citrate-stabilized GNPs were prepared by thermal reduction of $HAuCl_4$ with sodium citrate.[1] The following sequences of DNA oligonucleotides were used in this work:

Name	Sequences(5'to3')	Modified	Base number
H	CGTAGGAAAGACAGGTTGGAATCGTAGC	5'-SH$(PEG)_3$	28
G	GATTCCAACCTGTCTTTC	5'-SH$(PEG)_3$	18
I_A	CTGTCTTTCCTACG		14
I_B	GCTACGATTCCAAC		14
N1	CAACCTGT		8
N2	CCAACCTGTC		10
N3	TCCAACCTGTCT		12
N4	TTCCAACCTGTCTT		14
N5	ATTCCAACCTGTCTTT		16
M1	CAACCTGTCT		10
M2	CAACCTGTCTT		11
M3	CAACCTGTCTTT		12
M4	CAACCTGTCTTTC		13
M5	CAACCTGTCTTTCC		14

Equipment

Uv-Vis absorption spectrometer: Shimadzu UV-2550
micro-Raman spectrometer: Jobin Yvon Horiba HR 800 , with He–Ne laser(excitation line 632.8 nm), objective 50×.

Preparation of GNPs labeled with Raman dye

A freshly prepared Raman dye solution (0.4 μL malachite green or 0.8 μL cresyl violet at 1 mM concentration) was added dropwise to a rapidly mixing 1 mL GNPs respectively, which facilitated even distribution of the dye molecules on the gold particle surface. Higher concentration of dye molecule would made the nanoparticles prone to aggregation when negatively charged DNA in salt buffer was introduced to the gold-dye complex.

Preparation of GNPs Modified with PEG-thiol oligonucleotides

Single strand thiol-DNA was bound to the gold-dye complex following the procedure of Mirkin and workers with little modification. TCEP was used to reduce the disulfide bond of pegylated thiol-modified single strand DNA obtained from Sangon Ltd. The mixture of DNA and TCEP was stirred at room temperature for 120 min. Freshly prepared thiol-PEG-functionalized oligonucleotides were added to 1 mL gold nanoparticles suspended in PB-T (pH 7.4, 10 mM phosphate, 0.05% Tween-20) with final concentration of 100 nM. After 12 hrs, aqueous 2 M NaCl was added to the solution to bring the total NaCl concentration of the probe solution to 0.05 M, after standing for 4 hrs, the NaCl concentration of the probe solution increased to 0.1 M. Upon aging in 0.1 M salt for an additional 20 hrs, the nanoparticles were isolated by centrifugation 4500 rpm for 15 min with glass tube and washed three times with PBS-T (pH 7.4, 10 mM phosphate, 200 mM NaCl, 0.05% Tween-20).

Detect the outputs of DNA-GNPs based machine

In a typical experiment, 1 μL solution containing protector DNA (5 μM) in the single-stranded form was added to 50 μL host GNPs (PBS-T buffer) solution to initiate the assay. The system was allowed to gentle vortex over night. After washed with PBS-T,1 μL solution containing input DNA was added to the prepared host GNPs for another 2 hours. The guest GNPs probes (50 μL, PBS-T buffer) were then added to the solution and allowed to hybridize with gentle vortex for 12hours. After shaking by a vortex, solution was tested by UV-2550 or JY HR 800.

The influence of protector length in symmetric structure

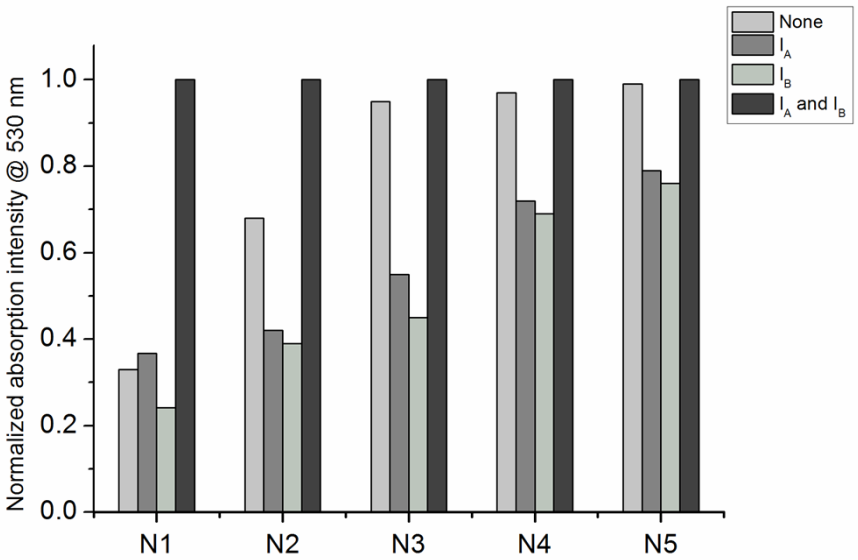

Fig. S1 As the length of protector increased from 8 to 16 nt (N1 to N5), the sealing effect (None) is increased. However, when the length is more than 12 nt, the extent of aggregation is lower (< 30%). Based on these findings, a twelve nt N3 was used in the symmetric structure that provided both sealing effect to guest GNPs and high extent of reaction.

The influence of protector length in asymmetric structure

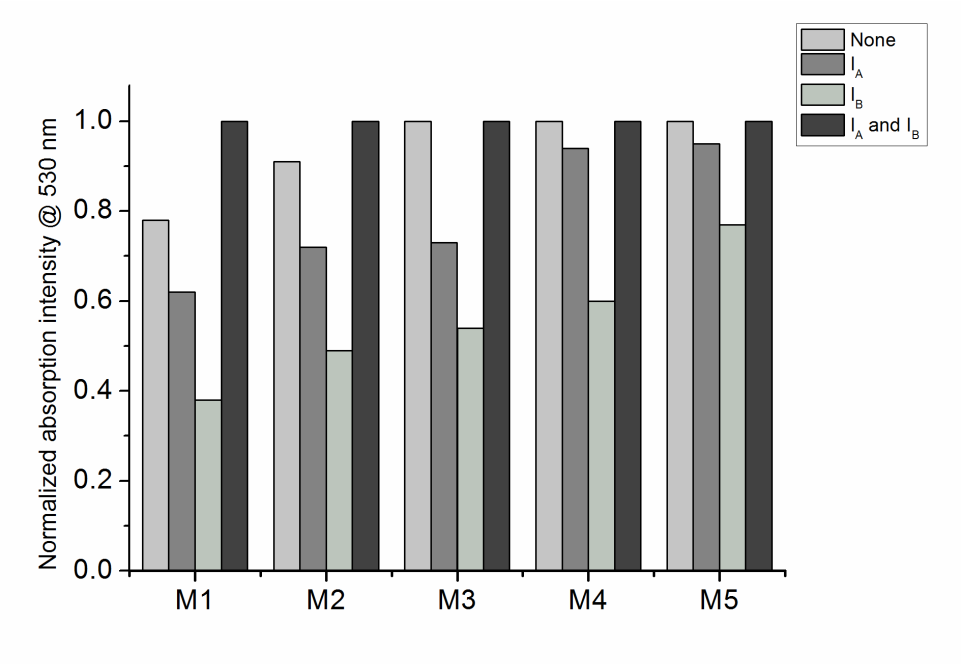

Fig. S2 As the length of protector increased from 10 to 14 nt (M1 to M5), the sealing effect (None) is increased. When the length is 12 nt (M3), both the extent of aggregation and sealing effect are acceptable. However, I_A is still able to trigger the aggregation until 13 nt M4 was used. Based on these findings, M4 was used in the asymmetric structure.

The influence of salt concentration

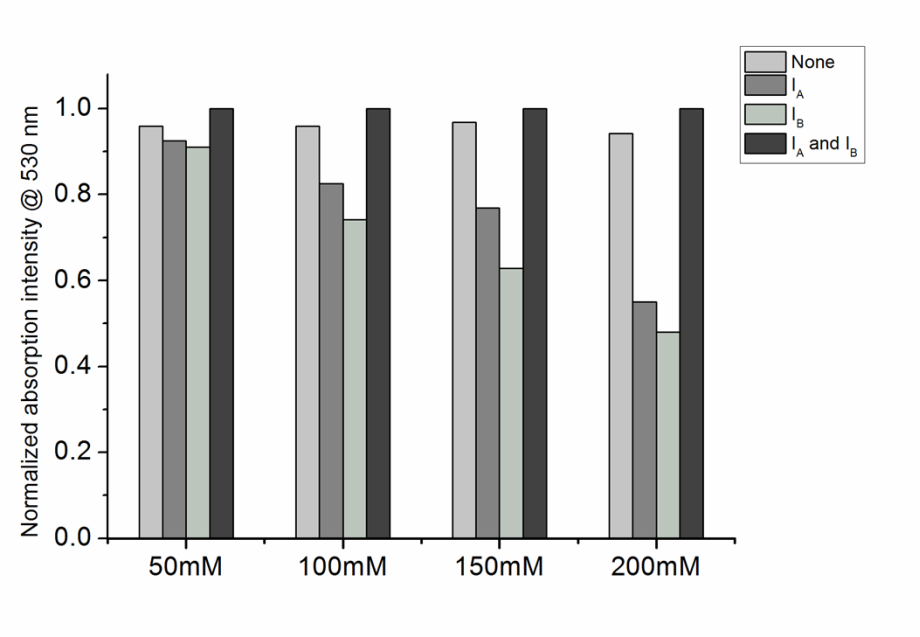

Fig. S3 The influence of salt concentration to symmetric structure DNA system. When the concentration of NaCl added into the solution increased to 200 mM, the DNA−GNPs showed high stability at (None) and (I_A and I_B).And have evident aggregation extent at (I_A) and (I_B) comparable to that of the lower salinity.

The SEM image of GNPs

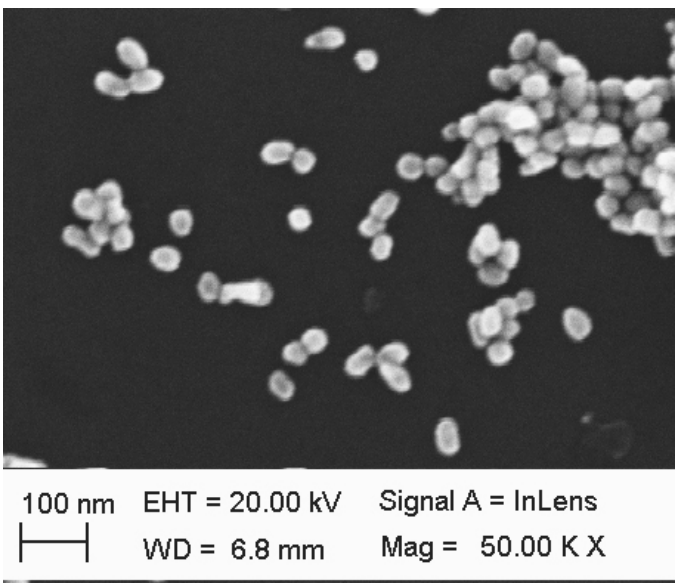

Fig. S4 The SEM image of GNPs. The mean diameter of GNPs is 35 nm.

The reproducibility of SERS

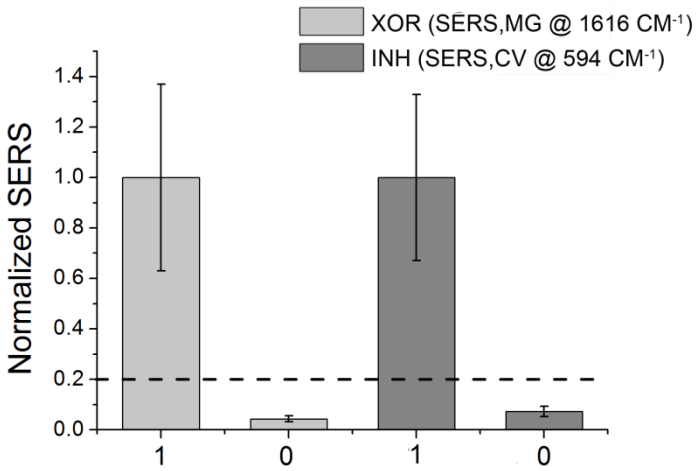

Fig. S5 The reproducibility of SERS spectra. Statistic results of the normalized Raman intensity at 1616 cm^{-1}(XOR gate) and 594 cm^{-1} (INH gate) on output 1 state and 0 state. Each bar are the statistical result from 13 record, respectively.

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